



Social behaviour and sociability traits of dairy calves raised in a cow-calf contact system

Cow-calf together project

Socialt beteende och sällskaplighet hos mjölkkalvar som är uppfödda i ett ko-kalv kontaktsystem

Maria Mwebaza Nabukalu

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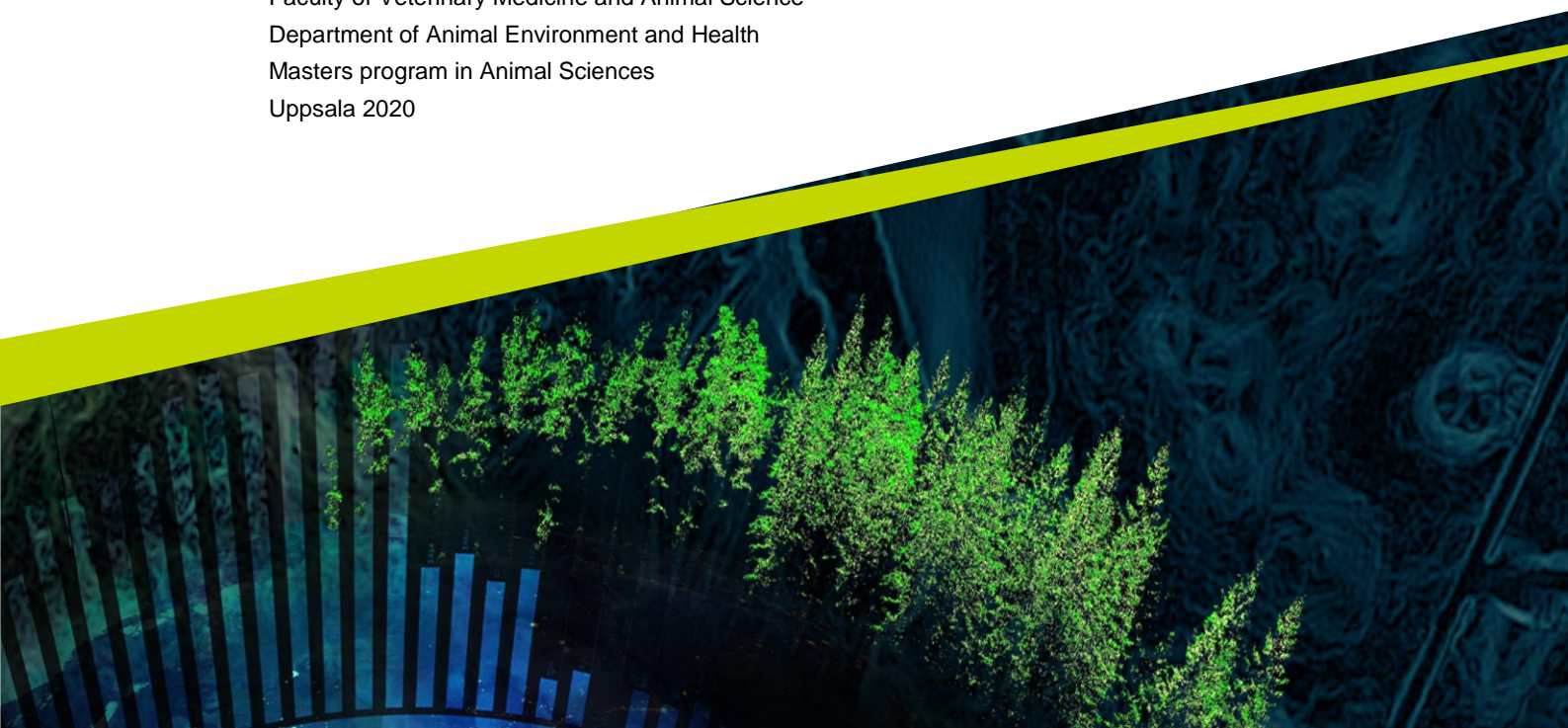
Swedish University of Agricultural Sciences, SLU

Faculty of Veterinary Medicine and Animal Science

Department of Animal Environment and Health

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Abstract

Calves are usually separated from their dams a few hours after birth, placed in individual house pens, cages or hutches and fed on milk replacer. This system has many welfare and ethical implications on the dam and calf, especially as regards to their bonding and natural behaviours.

A case-control behavioural observational study was therefore carried out to evaluate the potential effects of early socialization of calves raised with their dams, other cows, and peers during the first months of life, on later social behaviour and sociability traits in dairy calves. We investigated this on 23 dairy calves that were born in the summer of 2019 and subjected them to two treatments: Cow-Calf Contact treatment (CCC) (10 dairy calves) and Control group (C) (13 dairy calves). When the CCC and the C calves were 25 and 22 weeks of age respectively, video recordings and social behaviours of the calves using focal scans and continuous behavioural observations were done for at least 6 hours/day for five days; three consecutive days when calves were in their groups, one day when the groups were mixed and a day after mixing the groups. A week later when the CCC and the C calves were 26 and 23 weeks old respectively, a runway test was done to evaluate the calves' sociability. In addition, an avoidance distance test to evaluate animals' fear towards humans and to quantify the human-animal relationship were also applied. The effect of Treatments (CCC and C), Sex (heifer and castrated male calves) and their interaction were analysed in response to the observed behaviours of calves as the dependent variables. Weaning weights of the calves were included in the model as covariates to social and locomotor play behaviours. Before mixing, castrated males were walking ($F=5.76$, $P=0.028$) and self-grooming ($F=8.55$, $P=0.009$) more than the heifers. CCC calves performed more bucking ($F=5.91$, $P=0.026$) and jumping ($F=5.04$, $P=0.038$) locomotor play behaviours than C calves. Social motivation to socialize was observed more in the CCC calves through initiating interactions by performing more pushing, butting and mock fighting play behaviours when mixed. CCC calves expressed a higher motivation to reunite at latency <60s and spent more time in the zone closer to the grouped calves compared to the C calves which could indicate a higher motivation to socialize. Potential sociality for both CCC and C calves in the presence of humans was tested through their docility with an attained mean avoidance or flight distance of 0.9m in both treatment groups. CCC calves chin pressed ($F=7.40$, $P=0.022$) more than the C calves during the feed competition test. Therefore, raising dairy calves in social contact with their dams, other cows and peers is most likely to positively influence the early development and performance of natural behaviours. Additionally, it could potentially influence the acquiring and retention of sociability traits in calves to positively interact with both familiar and unfamiliar mates, which in turn could ensure animal welfare and well-being at a later stage in life when dairy calves are grouped.

Keywords: cow-calf contact system, animal welfare, sociality, sociability, socialization, dairy calves, affiliative and antagonistic behaviours.

Sammanfattning

Kalvar separeras vanligtvis från sina mammor några timmar efter födseln, placeras i enskilda boxar, eller kalvhyddor och matas med mjölkersättning. Detta system har många välfärds- och etiska konsekvenser för kon och kalven, särskilt när det gäller deras bindning till varandra och naturliga beteenden.

En fallkontrollstudie genomfördes därför för att utvärdera de potentiella effekterna av tidig socialisering av kalvar som uppfostrats med sina mammor, andra kor och kalvar under de första levnads månaderna, på senare socialt beteende och sällskaplighet hos mjölkraskalvar. Vi undersökte detta på 23 mjölkraskalvar som föddes sommaren 2019 och delades i två grupper som utsattes för olika behandlingar: Ko-kalvkontaktbehandling (CCC) (10 kalvar) och Kontrollbehandling (C) (13 kalvar). När CCC- och C-kalvarna var 25 respektive 22 veckor gamla, gjordes observationer och videoinspelningar av sociala beteenden hos kalvarna med hjälp av fokalskanningar och kontinuerliga beteendeobservationer i minst 6 timmar / dag i fem dagar; tre dagar i följd när kalvar var i sina behandlingsgrupper, en dag när grupperna blandades och en dag efter att grupperna hade blandats. En vecka senare när CCC- och C-kalvarna var 26 respektive 23 veckor gjordes ett sk. "run-way test" för att utvärdera kalvarnas sällskaplighet. Dessutom användes ett sk. "aviodance test" för att utvärdera djurs rädsla gentemot människor och för att kvantifiera förhållandet mellan människa och djur. Effekten av behandlingar (CCC och C), kön (kviga och kastrerade tjurar) och deras interaktion analyserades för kalvarnas beteenden. Kalvarnas avvänjningsvikter inkluderades i modellen som kovariat för sociala och aktiva beteenden. Innan grupperna blandades gick kvigor mer ($F=5.76$, $P=0.028$) och putsade sig mer ($F=8.55$, $P=0.009$) än de kastrerade tjurarna. CCC-kalvar bockade mer ($F=5.91$, $P=0.026$) och hoppande mer ($F=5.04$, $P=0.038$) än C-kalvar. Social motivation för att umgås med andra kalvar observerades mer i CCC-kalvarna genom att de initierade fler interaktioner när grupperna blandades. CCC-kalvar uttryckte en högre motivation att återförenas vid "run-way testet" och tillbringade mer tid i den delen av arenan som var nära de grupperade kalvarna jämfört med C-kalvarna. Det var ingen skillnad mellan CCC-kalvar och C-kalvar i deras rädsla för människor då det genomsnittliga flyktavståndet i "aviodance test" var 0,9 m för båda grupperna. CCC-kalvar pressade hakan mer mot andra kalvar ($F=7.40$, $P=0.022$) än C-kalvarna under foderkonkurrens. Att öka den sociala kontakten mellan mjölkraskalvar, deras mammor, andra kalvar och kor påverkar troligen den tidiga utvecklingen av naturliga sociala beteenden positivt. Dessutom kan det potentiellt påverka mer sällskapliga egenskaper hos kalvar, vilket i sin tur kan säkerställa djurens välbefinnande i ett senare skede i livet även när mjölkraskalvar grupperas.

Nyckelord: Ko-kalvkontaktsystem, Djurskydd, sällskaplighet, socialisering, tillhörande och antagonistiskt beteende, mjölkkalvar.

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Abbreviations

ADT	Avoidance distance test
AMS	Automatic Milking System
ANOVA	Analysis of Variance
C	Control system
CCC	Cow-Calf Contact system
GLMM	General Linear Mixed Model
HAR	Human-animal relationship
SEM	Standard error of the mean
SH	Swedish Holstein
SRB	Swedish Red Brown
VMS	Voluntary Milking System

1. Introduction

Dairy cows' welfare has mainly been centred on health and milk production with little concern on natural behaviours developed from the bonding of cow and calf through nursing and staying together (Ventura et al., 2013). The separation of newborn dairy calves from their dams a few hours after birth has incited concerns from members of the public (Boogaard et al., 2010). This is because, early separation of calf from its dam is often perceived to be unnatural and problematic for the welfare of the cow and the calf (Ventura et al., 2013).

Naturalness has typically been shown to be a lower priority for those working within agriculture especially dairy farming, who tend to instead emphasize concerns related to animal health (Te Velde et al., 2002; Vanhonacker et al., 2008), although this is not uniformly the case. Take for example some farms that have adopted alternative management systems that allow some contact between the cow and calf. This can be seen in European organic livestock producers who view naturalness as an important component of animal care (Lund et al., 2004; Vetouli et al., 2012; Spooner et al., 2012). In Sweden, Denmark, and Norway, it is mandatory to let the calf nurse at minimum 1-3 days after birth (Vidensenteret for økologisk landbrug, 2012; Debio, 2005; KRAV, 2012). This has led to alternative systems where calves are raised in contact with their dams for an extended period to be used hence increasing the interest of different stakeholders (Busch et al., 2017; Beaver et al., 2019). Stake holders like; scientists, producers, and consumers have developed increasing concerns and interest in the use of cow-calf contact systems (Brombin et al., 2019) especially on how the cow and calf are kept and the effect on their early life (Busch et al., 2017).

Therefore, this paper serves to study and identify the effects of early socialization on social behaviours and sociability traits developed and maintained by weaned dairy calves that were raised in contact with their dams, other cows, and peers. The study involves a comparison of social behaviours and sociability traits developed between two groups of calves. One raised with contact from birth until weaning when they were grouped, and another with calves that were separated a few hours after birth and raised in individual pens and later grouped after weaning. The purpose was to evaluate and identify which group of animals behaved socially better when the two groups were mixed to form a new combined group.

1.1. Popular scientific summary

Food security is best achieved through productivity, good welfare, and proper management systems of food animals. Recommended proper management and production systems should ensure good products like milk and meat that are from healthy animals for people to consume to be healthy.

Dairy calves require to be raised with their dams (mothers) and other cows to bond and quickly learn how to play, interact, and socialise. This early social environment can ensure the development and better performance of natural behaviour required for proper welfare, well-being, and probably maximum output in dairy production farms.

1.2. Aim

The aim of the study was to evaluate the potential effects of early socialization of dairy calves with their dams and peers during the first months of life, on later social behaviours and on sociability traits in the dairy calves.

1.3. Hypothesis

I hypothesized that:

H0: There is no difference in the social behaviours expressed by the cow-calf-contact calves and the control calves when socially challenged.

H1: The cow-calf-contact calves express more or higher sociability and proper play behaviours than control calves when socially challenged.

H2: The control calves express more or higher sociability and proper play behaviours than the cow-calf-contact calves when socially challenged.

2. Literature review

In modern dairy farming, calves are separated from the cows shortly after birth (within 24 hours post-partum) into group-housed pens or individually housed (single pens) and hutches. Despite the fact that individual housing protects calves from pathogens and makes it easier to monitor milk intake and health of the calves, the separation of calves from their mothers and away from peers deprives the calves of ample milk intake and bonding between cow and calf (Keeling and Gonyou, 2001).

Early separation is often perceived to be unnatural and problematic for the welfare of the cow and calf (Ventura et al., 2013). This is because, raising the calf together with the cow after birth has been proven to provide health and welfare benefits (Krohn, 2001; Flower and Weary, 2003). For example, calves reared with the dam have been proven to perform actively better, to learn or behave faster, like standing earlier after birth than those separated from their dams (Lidfors, 1996). It has further been proven that calves raised with cows gain more weight than calves reared artificially with restricted milk allowance (Flower and Weary, 2001), and show reduced signs of distress during an isolation test (Duve et al., 2012). Cow and calf together additionally ensure natural nursing behaviours of dams which maintains proper udder health of a cow hence supporting bonding, and increases social abilities in calves (Johnsen et al., 2016). The cow-calf contact system is further assumed to have the potential to improve animal welfare through improved behavioural, social, and physiological adaptability (Buchli et al., 2017). Social learning of feeding for example sampling of solid feed in the company of grazing adults in the first few weeks of life (Key and MacIver, 1980; Nolte et al., 1990), decreased food neophobia and acceptance of novel foods (Lynch et al., 1983; Galef and Stein, 1985) are all learnt by calves raised with dams and other cows which may be particularly beneficial before and after weaning. This is particularly beneficial to the calves in the period when they make the transition from a milk-based diet to solid feed (De Paula Viera et al., 2012).

Cow-calf systems that allow calves to stay with dams are feasible, and long-term positive effects of calves staying longer with the cow can be seen when heifers are introduced into the dairy herd (Wagner et al., 2012). Keeping calves with their peers and with dams for longer nursing stimulates affiliative and natural behaviours which contribute to a normal individual and social interactive behavioural

development of a calf (Shamay et al., 2005). Longer nursing additionally ensures higher weight gains for the free suckling calves with a recorded average weight gain of 0.9kg to 1.4kg (Grondahl et al., 2007; Roth et al., 2009) than those reared without the dams which are 0.5kg to 0.6kg per day (Mcgee et al., 2018). The increased milk intake by the calf through suckling from the dam (ad libitum feeding) probably impacts on a high pre-weaning calf weight gain which in the later growth stage contributes to higher milk yields especially at the heifer's first lactation (Shamay et al., 2005).

Despite the positive attributes, negative implications of low milk production have been reported in the cow-calf-contact system. A depressing weight gain of calves raised with dams at weaning if not well handled and managed at the separation of calf from the dam due to stress is one of them (Johnsen et al., 2016). However, proper weaning methods can be devised; calves housed with the dam but prevented from suckling have been associated with higher weight gain than calves given the same amount of milk but isolated from the dam (Krohn and Colleges., 1999). This shows that the maternal presence of the dam is very important and may have a positive effect on weight gain independent of milk transfer. Hence, justifying the feasibility of contact of a calf with a cow and its peers even without nursing.

2.1. Early social environment and grouping of calves

Separation of calves from dams and a lack of early social experience alters the current and influences the later social development of calves and hence behaviour especially when mixing unfamiliar individuals (Boe and Faerevik., 2003). Proper early social environment has, therefore, been ensured by the EU-directives by putting up protocols that enhance good welfare. EU-directives have also ensured that appropriate social behaviours and interaction of calves are achieved through, visual contact of calves raised in single housing pens or hutches facing each other for social contact (Bach et al., 2009; Rushen et al., 2008; Jordbruøksverket, 2014). Hutches or individual house pens are meant to face each other so that the calves can have a clear vision of other peers and that calves older than eight weeks are housed in groups (Jordbruøksverket, 2014). The directives show the importance of social grouping and interactions which could directly influence the behaviours and proper development as well as well-being and health of dairy calves.

In socially challenged environments, group-reared calves have a higher rank than individually reared calves when housed together (Broom and Leaver, 1978). Calves reared in single pens where social contact with peers is limited to head contact in front of the pens, showed more fear in a novel environment and a higher heart rate with a longer latency to sniff unfamiliar calves than group-reared calves at three

months of age, but not 6 months of age (Jensen et al., 1999). Additionally, more affiliative behaviours and fewer agonistic behaviours have been observed among calves previously kept in individual crates, pens or hutches compared to calves reared in groups (Vaissier et al., 1994). Hence, the grouping of familiar calves together is important as it facilitates early development of social responses in calves as they exhibit a lot of agonistic play behaviours (Jensen et al., 1999). Early social environments also facilitate the development of mutual social attachment that are relatively long-lasting and would survive temporary separation (Keeling and Gonyou., 2001).

2.2. Socialization and Sociability

Socialization is the process of acquiring social rules and adopting behavioural patterns applicable to an individual's social environment (Petak I., 2018). Through a process, an animal develops its own species' identity and personality hence learning how to interact socially with its own species and other living creatures of different species or breeds (Petak I., 2018). Animals, therefore, learn to read or perform different behaviours and hence learn to have good social manners through interaction. Socialization starts in young animals and it is the most effective and sensitive period before maturity, but it is a lifelong process (Petak I., 2018).

Environment and upbringing of an individual animal influences the social development and life of that individual while social contact improves the social skills and stimulates solid feed intake of the animal (Keeling and Gonyou., 2001). Socialization or sociality and sociability are all aspects of social motivation. Sociality of an animal is the extent to which it needs social companionship while sociability is how close an animal wants to be close to other group members or how frequently the animal interacts with other group members (Keeling and Gonyou., 2001).

Vocalization has been assessed in relation to sociality by recording vocalisation when calves are in social isolation as done by Syme, 1981; Faure et al., 1983 and in the 'treadmill test' (Mills and Faure, 1990). Literature has shown that socially housed calves vocalise less than individually housed calves when they are weaned (De Paula Viera et al., 2010) suggesting that the presence of a peer provides a buffer during stressful management procedures and conditions (Jensen et al., 2015). However, Fraser and Broom (1997), states that vocalization is a crucial communication behaviour or language for animals, and it can be a sign or alarm for distress which explains why socially housed calves highly vocalize when separated from their peers or group.

2.2.1. Sociality traits

Social play includes play fighting (like pushing, mock-fighting, butting) which involves interactions that do not result in flight or submission and non-reproductive mounting which are often performed in connection with locomotor play (like bucking, jumping, galloping, running) (Jensen et al., 1998).

Social traits are normally known or described as vigorous play behaviours for calves, and they tend to require space and social facilitation (Reinhardt et al., 1978; Reinhardt and Reinhardt, 1982). Social interactions between free-ranging calves have been reported to increase over the first few weeks of life (Wood-Gush et al., 1984; Vitale et al., 1986; Kerr and Wood-Gush et al., 1987) with less reported vocalizing in socially housed calves than individually housed calves when they are weaned (De Paula Vieira et al., 2010).

Studies in semi-wild ungulates suggests that social play increases during the first few months of life (Vitale et al., 1986), and the decline observed in Jensen et al., (2015) was a transient due to weaning.

Sociality can be identified through the docility and human-animal relationship (HAR) of the calf.

HAR is described as the mutual perception of the animal and the human, based on previous interactions, and as reflected in their mutual behaviour (Waiblinger et al., 2006; de Oliveira et al., 2020).

3. Materials and methods

3.1. General aspects

3.1.1. Ethics

This experiment was conducted in compliance with the animal ethics application Dnr 5.8.18-18138/2019 at the Swedish Livestock Research Centre Lövsta lantbruksforskning, SLU located in Uppsala, Sweden.

3.1.2. Performance

Body weights for all the calves were measured and registered weekly to monitor their weight gain from the day they were born until the calves were 56 and 112 days old for the C group and CCC group, respectively. On average, CCC calves gained 1,19kgs/day while the C calves gained 1,08kgs/day from when they were born until before the study was carried out.

All animals appeared healthy while the experiment was being carried out and there was neither an accident nor severe illness on any of the calves except for some minor coughing from a few calves that were still reported to be in a good body and medical condition.

3.2. Animals and housing

The experiment was carried out at the Swedish Livestock Research Centre, Lövsta located in Uppsala, Sweden over a period of one month in February 2020. Twenty-three dairy calves (13 heifers and 10 castrated male calves) born in the summer of 2019 were used in the experiment of which, fourteen of the calves were of the breed Swedish red brown (Srb) and the remaining ten calves were of the breed Swedish Holstein (Sh).

When the experiment began, the average age for the C group was 146 days, mean birth weight (BW) was 39kgs and mean weaning weight was 196.9kgs. The eldest

C calf was born on the 2nd of September and the youngest on the 25th of September 2019.

For the CCC calves, the average age of the calves when the experiment started was 165 days, mean BW was 39.2kgs and the mean weaning weight was 236.3 kgs. The eldest CCC test calf was born on the 14th of August 2019 and the youngest on the 10th of September 2019.

Calves were randomly divided into two groups: The Control (C) group (13 calves; =6 castrated males, 7 heifers) and the Cow-Calf Contact (CCC) group (10 calves; =4 castrated males, 6 heifers) as shown in Table 1, below

Table 1. Distribution of the calves used in the experiment and their details

Treatment	Heifers (%)	Castrated Bull calf (%)	SRB (%)	SH (%)	Mean weight (kg)	Mean age (days)
C	53.8	46.2	61.5	38.5	196.9	146
CCC	60	40	60	40	236.3	165

3.2.1. Control group

All C calves suckled colostrum from their dams within the first two hours after birth. After colostrum was consumed, the calves were individually housed in calf pens, fed on 6L of whole milk per day divided into 3+3 litres (one serving in the morning and one other in the afternoon). Milk was administered to the calves using buckets with teats that were supplied with milk from a milk tank using a “milk taxi” that ensures appropriate temperature of the milk. Calves were weaned at 56 days of age after birth and then moved to a group pen in the calf barn. They were all managed according to the standard routines of the Lövsta herds and all of them were healthy without any birth defects.

3.2.2. Cow-calf contact group

A cow-calf contact (CCC) can be described as one where there is physical contact between a dam and her own calf, or between a foster cow and her foster calf (Sirovnik et al., 2020).

All CCC calves suckled colostrum from their dams in the first two hours after birth and then kept in the calving pen with the cow for the first 2-3 days post-partum. In the CCC system, cows were fetched from the calving pen daily to be milked in the Voluntary Milking System (VMS) unit. After the first 2-3 days, the cow-calf pair were introduced to a group of cows. Five of the calves and their cows were introduced to the indoor/VMS group and the other five calves and their cows were introduced to the outdoor/pasture group.

The VMS/indoor group (Figure 1) was a contact area where calves stayed all the time. Cows had access to the contact area where they could meet their calves, access cubicles, concentrate feeders and water. At the far end of the contact area was a calf creep which had cubicles for only the calves' access. A passage selection gate that could only be opened by cows directed the cow to the Automatic Milking System (AMS) for milking in the VMS-unit. After the milking, the cow could access a selection gate back to the contact area.



Figure 1. "Indoor contact area" Photo by: Singrid Agenäs, SLU

The outdoor group (Figure 2) was a contact area in a pasture pen made with a mobile shed (Figure 3) consisting of a calf creep and a place where cows and calves had access to water, hay, and concentrate. There was also a scale to measure body weight gain. The cows could leave the contact area through a one-way selection gate (same as the in-door set-up) and they could go to the VMS unit where they had access to comfort brushes, the milking unit (AMS), roughage, concentrate and other cows.

At 56 days of age, cow-calf contact was switched to half day and all CCC calves had access to the cows only during the daytime until they were 112 days old. Calves were separated from their mothers with a gate at night (for 12 hours) so, cows and calves could only see each other to make contact during the day and calves could suckle in the 12hour half day contact. At 112 days of age when the calves were developing puberty signs and weighing roughly 200kgs, they were abruptly weaned, separated and removed from the dairy unit into a group of CCC calves originating from the two contact areas as shown in figure 1 and 2.



Figure 2. "Outdoor contact area" Photo by: Singrid Agenäs, SLU



Figure 3. Mobile shed from playmek (www.playmek.se/mobilt-vindskydd) Photo by: Mia Jernhake, SLU

3.3. Experimental set-up

A parallel-group design involving the study of two different treatments simultaneously was done as recommended by de Oliveira et al. (2020). This was done to avoid confounding effects such as season, load of infectious diseases or staff changes (de Oliveira et al., 2020). C calves were raised in a normal farm regulated structured system without being exposed to any special treatment and the CCC calves were raised with their dams, other peers, and cows' contact through two treatment systems. The C and CCC calves were group housed at 112 and 147 days of age respectively after weaning and placed in adjacent pens separated by metal barriers. Each pen measured 8.73m x 5m as shown in figure 4 despite the difference in the stocking rate (C=13; CCC=10) between the two groups. Therefore, space allowance was 3.36m²/calf, for the C group and 4.37m²/calf for the CCC treatment group. It is evident that there was a 1m² space allowance per calf difference between the C and CCC treatment calves that could potentially be raised

as a limiting factor in space allowance for play behaviour. However, both groups received a space allowance per calf that surpasses the recommended 1.5 to 2m² space allowance per calf (Jordbruksverket, 2014) and there might not be an impact on the social interaction since both groups performed play behaviours so space could be considered a non-limiting factor.

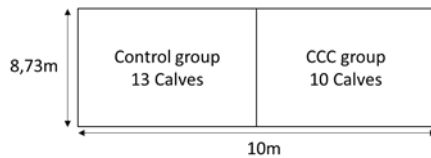


Figure 4. Holding area/ house pen of experimental calves

Calves were fed ad libitum roughage and concentrate feed by the farm staff and clean running water was provided daily from drinking bowls throughout the experiment. All calf housing requirements and conditions like light, temperature, humidity, housing, and air circulation were met according to the Swedish animal rights, regulations, and protocols. All calves that participated in the experiment were healthy and in good body condition.

3.4. Experimental designs and procedures

3.4.1. Marking of the calves

The calves were tagged with four-digit tag numbers at birth, and these were used as identification numbers for each calf at the farm. For simple identification during the behavioural observations, calves were marked randomly and by chance with different colour collars (as shown in the appendix). The allocated colour collar for each calf was recorded in sync with each calf's tag number to be used while identifying calves in the experimental tests and during the data analysis.

3.4.2. Behavioural observations

These were carried out to identify the agonistic and affiliative interactions, social skills, and personality traits of dairy calves from the two different test groups exposed to two different early environments during their social encounters. To ensure that the interpretation of the protocols and behaviours between the different observers were accurate, the three observers had a one-day practice session together in the first week of February 2020 before the start of the study. The behaviours recorded by all observers for each session and within the treatment groups were compared to ensure that observers made the same interpretation of the protocols and behaviours. Any observed behaviours missing in the protocol that was

identified by all observers in the test sessions were added to an updated protocol to be used in the study.

In the second week of February 2020, the behavioural observations begun, and they were done for five days when the calves were 147 and 161 days of age for the C and the CCC calves, respectively.

Calf treatment groups were housed adjacent to each other in the calf barn where they were grouped housed after weaning, between November and early December 2019, and throughout the entire experiment in February 2020 (Details in Table 2).

Table 2. The mixing of the calves, the observations and all behavioural tests were done on the same time but the calves in the two treatments were at different ages

Time in age	Control calves	Cow-Calf-Contact calves
Birth	Separated after 2 h post-partum	Kept with cow in pen 2-3 days
0-56 days	Kept individually and fed whole milk using a bucket with teats.	Kept in group with cows and calves, suckle mother
56-112 days	Weaned and group housed	Kept in group with cows and calves, suckle only during the daytime (12hr/day)
112-140 days	Group housed	Abruptly weaned and separated from cows at 16 weeks, then group housed
140 days	Preparation for experiment, practice, and habituation of animals to handlers' presence.	Housed in a group pen consisting of 2 differently treated groups of calves.
147 days	Social Behaviour observations and data collection (3 days) and Habituation to test arena/corridor	Group housed
154 days	Habituation to experimental test arena (2 days) Runway test (2 days) and Avoidance distance test (one day).	Preparation for experiment, Practice, and habituation of animals to handlers' presence.
161 days	Mixing of groups (One day: 24 th of Feb), Social behaviour data collection (2 days) and Feed competition test (One day)	Social Behaviour observations and data collection (3 days) and Habituation to test arena/corridor
168 days		Habituation to experimental test arena (2 days) Runway test (2 days) and Avoidance distance test (one day).
175 days		Mixing of groups (One day: 24 th of Feb), Social behaviour data

Behavioural observations were done in five days starting with three observation days in the second week of February 2020 when the calves were in their treatment groups aged 147 and 161 days for the C and CCC calves, respectively. The remaining two behavioural observation days were done in the fourth week of February 2020, on the mixing day and on the day after mixing the two treatment groups when the calves were aged 161 and 175 days for the C and CCC calves respectively (Table 2). Behavioural observations lasted 6hrs/day in all the five observation days. For the first three observation days (before mixing the calf groups) and on the 5th day (the day after mixing groups), the observations done on each day started with a focal scan for five minutes with a state observation of the posture or position and behaviour of each calf being recorded followed by a 40 minute continuous social observation where social and individual play behaviours were recorded based on the ethogram described in Table 3.

Table 3. Ethogram used for the observations in the control and cow-calf-contact groups

Observation	Type	Level	Type	Description
Standing	I	Posture	State	Upright and body weight supported on three or four of the hooves which should be in contact with bedding.
Lying	I	Posture	State	Sides or entire stomach and body in contact with ground, object or bedding without the hooves bearing the body weight. In the process of lying down or standing up.
Walking	I	Posture	State	Shift of body weight from one location to another with all four hooves.
Vocalization	I	Behaviour	Event	Vocal sound produced either unintentionally or intentionally following an activity or presence of an individual.
Eating	I	Behaviour	Event	Picking feed and chewing any form of eating material. Searching for feed while sniffing and digging with the muzzle <3cm from the feeding alley.
Drinking	I	Behaviour	Event	Mouth inserted in the drinker
Seek Interior	I	Behaviour	Event	Searching through or looking for something in space, straw, and interior of the pen
Self-groom	I	Behaviour	Event	Licking any part of own body or head while the tongue is in contact with the fur

					or scratching with hind leg lifted from the ground and muzzle is in contact with fur
Sniff interior	I		Behaviour	Event	Smelling or positioning the muzzle in an inch's contact with any object, material, or interior of the pen
Lick interior	I		Behaviour	Event	Tongue in contact with any object or material. In the process of tongue withdrawing or licking any object or material in the interior of the pen that is not feed.
Ruminate	I		Behaviour	Event	Moving jaws in a rhythmic manner or chewing / regurgitating when standing or lying away from the feeding area.
Rubbing	I	C ²	Behaviour	Event	Any body part in contact, applying friction or pressure to another.
Body Sniff		C ^{2,3}	Behaviour	Event	Muzzle in contact or within an inch's distance of contact with fur or a companion's body part
Genital sniff		C ³	Behaviour	Event	Muzzle in contact or in an inch's distance of contact with the genital area with a companion
Body lick		C ^{2,3}	Behaviour	Event	Tongue in contact with its own or companion's body or within one muzzle-width of the companion.
Genital lick		C ³	Behaviour	Event	Tongue in contact or within an inch's contact with the genital area of a companion
Push		C ^{2,3}	Behaviour	Event	Displacing another from one position to another by exerting force either with the head or any body part.
Butting Calf		C ^{2,3}	Behaviour	Event	A slight push, press or rubbing the head against the head or body of another calf.
Mock fight		C ^{2,3}	Behaviour	Event	Two calves Standing forehead to forehead while mutually pushing in the direction of each other (Duve & Jensen, 2012).
Mount		C	Behaviour	Event	Both forelegs are lifted from the ground, while jumping onto the back side of another calf while using the hind legs as support on the ground.

Nothing	I	Behaviour	Event	Staring into space or in an inactivity state of the body
Locomotor play	I	Behaviour	Event	<p>Jumping: Both forelegs are lifted from the ground and stretched forward might be possible to be followed by both hind legs being lifted from the ground.</p> <p>Bucking: Both hind legs are lifted from the ground and stretched backwards, maybe followed by the forelegs being lifted from the ground.</p> <p>Running²: Chasing companion (s) and rapid or continuous movement in circles, back and forth or with constant changes in direction.</p> <p>Butt fixture: Pushing the head with force against the wall, feed barriers, and water bowl or on the ground.</p> <p>Straw play: Swatting straw with one hoof while standing. Rubbing head, body, throat, or neck in the straw while lying or kneeling on both forelegs.</p>

¹I = Focal scan; C = Continuous recording. ²Calves in pairs performed the same activity at the same time while observing, which was recorded as mutual. ³Behaviours normally performed in sequence while observing.

During the observations, a calf was recorded with one of three mutually exclusive position behaviours, Standing, lying, and walking (Table 3). In addition, the activity of the calf was recorded as of either vocalizing, eating, ruminating, self-grooming, body licking/sniffing, genital licking/sniffing, licking/seeking interior or drinking independent on whether the calf was walking, standing, or lying. Observations within the course of the day were further divided into sessions done based on an intertwined focal scan and continuous observation schedule as shown in the appendix.

On the fourth observation day which was the mixing day, calves were mixed to make one large, combined group containing both C and CCC calves. Due to the high social interaction at the time of mixing, the observations started with a 60-minute continuous observation of social interactions followed by a 10-minute focal scan. The following observations all through the sessions in the day were based on a schedule as shown in the appendix and following the time interval of 40 and 10 minutes for continuous and focal scans, respectively.

Because of the transition from small groups to one large group on the day of mixing, three observers were involved in the behavioural observations and all were well

positioned each in the centre of the group assigned to be observed as shown in figure 5.

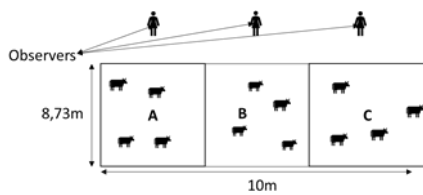


Figure 5. Divisions A, B, and C in the calf housing pen on the mixing day and after mixing the groups and respectively showing the allocated positions for the three observers 1,2 and 3 during the behavioural observations on the day of mixing.

Observers' allocated group of observation was done randomly at the beginning of each behavioural observation session in a day but the group to be observed next and in the course of the day was swapped and changed simultaneously in each new session to minimise the error, bias and ensure consistency.

3.5. Behavioural tests

3.5.1. Runway test

The test was done in the third week of February 2020 at ages 154 and 168 days for the C and CCC calves respectively, and that was before the treatment groups were mixed (Table 2). The test was based on a similar principle as the treadmill test (Mills and Faure, 1990) and the runway test (Gibbons et al., 2010). The purpose was to assess the social motivation and sociability by assessing the animals' latency to overcome a distance and reunite with the peers (de Oliveira et al., 2020). Animal's fear and social reinstatement responses to social isolation and sociality through vocalization, exploration and locomotion were tested (Wagner et al., 2015). The calves from the two treatment groups (C: n = 13, CCC: n = 10), were subjected to a runway test only once each in an experimental period of two days.

On day one, 12 randomly selected focal calves from both treatments were assigned to two groups (CCC calves: n=5, C calves: n=7) to avoid mixing them while carrying out the runway test. On day two, the remaining 11 focal calves were tested (CCC calves: n=5, C calves: n=6). The reason for testing calves from both treatments on each experimental day was to create a balance and avoid bias and stress exposure in the calves during the process of separation and movement of the same group in a short period of time which would in turn affect the experimental test and results.

The Runway (14.7mx1.76m) was a concrete floored corridor that was approximately 36metres away from the calves' home pen but located between the

feed storage and distribution area, walkway to the heifer housing pen and to the boots and cleaning corridor leading to the tearoom. The runway corridor was also used as a passage for handlers and animals being moved from one pen to another. The test calves had to be habituated to the area for two days before the runway test was done. Calves of each group were moved from their pen to the corridor as a group and left to walk around and get familiar with the flooring, environment and sound in the area in a systematic way with a 30 minutes interval per group for 4 rounds (two in the morning and two in the afternoon).

On the two test days, the test groups were penned at the furthest end of the runway (Figure 6) for 10 minutes so that they could relax and habituate to the area.

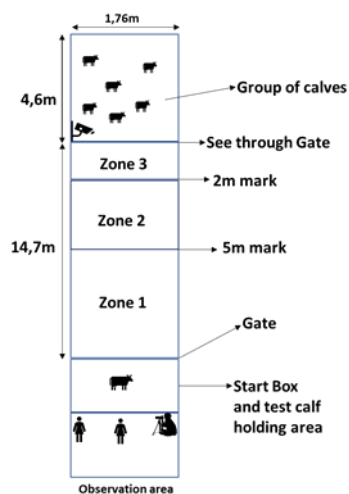


Figure 6. Lay out of the runway test arena.

In turn, each calf to be tested was removed from its test group and gently moved by three familiar experimenters to the start box located at the other end of the runway as shown in Figure 6. The calf was penned and locked behind a gate that was completely covered to cut off visual contact between the test calf and the test group and to allow it to settle and habituate there for a further 5 minutes before being tested. After the 5 minutes habituation in the holding pen, two experimenters released the calf, allowing it the freedom to move out of the holding pen and move up and down the passageway labelled with 2m and 5m marks for the runway test. The test duration was 300s from when the calf crossed the gate which is where the start line was marked. A calf was considered to have reached the 5m and 2 m marks when the front legs had crossed the marks. After the test, the test calf was marked for easy identification and separation from non-tested calves and then put back into the test group. The next calf to be tested was then selected and led through the same protocol for testing. Animals were selected in a random test order for ease of access without causing stress.

All test sessions were well timed and recorded using digital camcorders (Sony HDR – CX240E 3.6V, 9.2 Megapixels and Garmin VIRB Ultra 30 action camera) by well-trained experimenters who were blocked off by a completely enclosed gate in the observation areas as shown in figure 6 so that they could not be seen by the test calf and the other calves in the test group. The latency to reach the 5m and the 2m marks from the test group, and the duration of time spent in the 5m and 2m areas of the runway test were taken from the video recordings and used as measurements of social motivation.

3.5.2. Avoidance distance test

The avoidance distance test (ADT) test was done in the third week of February 2020 before mixing the two treatment groups which were aged 154 and 168 days for the C and CCC calves, respectively (Table 2). The purpose of carrying out this test was to test the sociality and the human-animal relationship (HAR) through the ability of the CCC calves in comparison to the C calves to interact with humans. The test was performed away from the housing barn of the test calves and following a stationary person test (de Oliveira et al., 2020)

The calves from the two treatment groups were subjected to an ADT carried out once per calf on either one of the two periods of time (morning or afternoon) of the scheduled experimental day.

On the experimental day, the test sessions were divided into two sessions. The morning session, which started at 09:30 am to 12:00 noon with 12 randomly selected focal calves (different random selection of test calves from the runway test) assigned to two groups (CCC calves: n=5, C calves: n=7) and the afternoon session which started at 01:00 pm to 03:30 pm consisting of the remaining 11 focal calves. The remaining 11 focal calves were tested in two groups (CCC calves: n=5, C calves: n=6) as the initial group in the morning. The test was done starting with the randomly selected calves from the C group followed by the CCC treatment group simultaneously to avoid mixing them while carrying out the test. Random selection of calves from each group and selection of one group from each treatment at a time and per session was done to balance and avoid stress amongst the calves in the process of separation and movement from their housing pen to the test arena.

The same test arena used and as described in the runway test was used for the avoidance distance test but with the testing area measuring 18m x 1.76m for this test. The animals were habituated to the arena before all the tests (as described in the runway test) since it was the same corridor used as the arena for both tests.

On the experimental day, two handlers participated. The test calves were penned at one end of the arena (4.6m x 1.76m, figure 7) for 10 minutes so that they could relax and habituate to the area. In turn, each calf to be tested was removed from the test group and gently moved by the two familiar experimenters through section B to the test area located at the other end of the arena (Figure 7).

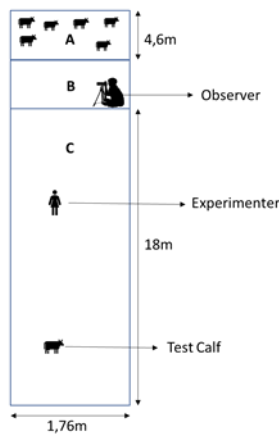


Figure 7. Lay out of the avoidance distance test arena.

The experiment involved two experimenters each of which had specific individual tasks during the test. One of the experimenters was, measuring the avoidance distance while carrying out the test on each test calf, standing in the arena section C (Figure 7). The other experimenter was timing, observing, and recording the measured avoidance distance of each test calf, while standing in the arena section B (Figure 7) and marking every tested calf at the end of each test.

During the ADT, one of the experimenters (placed in section C, Figure 7) ensured a clear visual contact between the calf and herself (that is, the calf should be looking at the experimenter), and then moved slowly in the direction of the calf while maintaining a visual contact until the experimenter reached one metre distance between the calf and herself. At the one metre distance, the experimenter then stretched out one hand towards the test calf's head at the muzzle level while the other hand supported at the waist was holding a laser distance meter (Biltema Art. 15-740). The experimenter then started to make gradual steps towards the calf to determine its receptive or approachable reaction towards her. The receptiveness or avoidance was determined by measuring the distance the calf either moved forward or jerked backwards respectively and the movement of the test calf's legs and body or turning of its head away from the experimenter.

Every test calf was locked behind a gate that was completely covered to cut off visual contact with the test group and the handler who was doing the observation and recordings to avoid any disruption of the experiment.

The test duration for each test calf was limited to a maximum of 60 seconds from the time a proper direct positioning at the one metre distance between the calf and experimenter was obtained. After the test, test calf was marked and returned to the test group and the next calf to be tested was then selected and led through the same protocol for testing. Animals were selected in a random order for ease of access by

ensuring that the calf nearer to the gate and not marked as tested was selected next for testing in order not to cause stress and injury to the calves in the group.

All test sessions were well timed and recorded using digital camcorders (Sony HDR – CX240E 3.6V, 9.2 Megapixels and Garmin VIRB Ultra 30 action camera) by the second researcher who was blocked off by a completely enclosed gate in section B (Figure 7).

3.5.3. Feed competition test

This was the last test done for the study in the fourth week of February 2020 when the calves were aged 161 and 175 days of age for the C and CCC calves respectively (Table 2). This test was done a day after the two calf groups were mixed. It was reciprocated based on similar principles as in the research paper by Duve et al., (2012). The purpose of this test was to assess the social skill of the calves from the different groups by identifying the threat or aggressive agonistic behaviours performed representing either the submissiveness or superiority of the calves when socially challenged.

After 48 hours in the mixed group pen, the test calves were subjected to a feed competition test in the same calf barn that the calves were housed. However, in the calf barn, the separating barriers between the two group pens were removed to make one large, combined pen where all the calves were maintained and later deprived of concentrate and hay for 16 hours (from 18:30 to 09:30 hrs). The purpose was to test the new formed group and evaluate how the two treatments affected the ability of the test calves when grouped to find and compete for feed in a novel setting.

Prior to the test day, the test calves had two feeding areas with each positioned at the extreme end of the fore front of the pen. Each feeding area had a feeding panel with seven upright rectangular openings available for the calves to reach the feed trough. Each rectangular opening was allowing room for only a head of one calf at a time. In the evening before the test day, hay and concentrate feeding was stopped. On the test day, only one feeding area (feed trough on the extreme left side of the pen) of the two feeding areas was chosen to be used during the test which was then replaced with a continuously open lined feeding panel that was not partitioned. The feeding panel could allow approximately eight calf heads in the feeding area at a go (Figure 8).



Figure 8. Feeding area during the feed competition test at Lövsta. Photo by Maria Mwebaza, SLU.

At 09:00 to 09:15 am of the test day, cameras (Sony HDR – CX240E 3.6V, 9.2 Megapixels and a Garmin VIRB Ultra 30 action camera) were fixed and tested to be at a good view of the feeding area so that they can monitor the behaviours of the calves during the test. At 09:20 to 09:30 am, the researchers provided a minimal amount of roughage that would ensure competition of the calves on the selected test feeding area during the test. At 09:30 am, the test begun with the researchers starting the recording using the well-placed cameras, giving access of roughage to the calves to feed, and leaving the test room with all the doors closed. However, it should be noted that calves were not locked up to prevent them from approaching the test feeding area and starting to eat before the test begun. Therefore, the calves that were standing at the test feeding area started eating immediately the feed was placed in the feeding area and before the test and recording started.

The test lasted 20 minutes and then the researchers opened the concentrate dispensers and placed more feed on both feeding areas so that the calves could feed.

3.6. Behavioural analysis

Behavioural video recordings that were done during the Runway test, and the Feed competition test were analysed by VLC 3.0.11 for Windows 64 bits with Boris software 7.9.8 version.

3.6.1. Runway test

Video recordings on 22 dairy calves were collected and a coded ethogram was developed based on essentially identified behaviours (Table 2). The ethogram was encoded and used in Boris to analyse the videos.

Due to a battery power shut down during the experiment, data for one calf from the CCC group was missing. That is why instead of 23 test calves, only 22 calf results were obtained from the video recordings transposed into the behavioural analysis excel sheet data.

Latency for calves to pass a given mark from the starting point to: 2m and 5m for zones 3 and zone 2 respectively was recorded.

A time interval and schedule were also developed using a time frame in seconds for the time it took calves to move from one zone to another (latency) and the duration recorded in seconds for the time each calf spent in each zone. The number of times a calf occupied a given zone was also recorded and all data was transposed into an excel sheet for further statistical analysis.

Vocalization of each calf while in isolation and habituating in the start box and during the runway test was recorded and transposed into an excel file for further analysis and interpretation.

3.6.2. Feed competition test

An ethogram based on eating behaviour (Table 2), was developed, and used in the Boris project software to analyse the calves that were observed eating at the feeding table at every 30 seconds interval. The results were transposed into an excel sheet for statistical analysis.

Behaviours from the ethogram in Table 2 were coded and used to analyse the recorded video for agonistic behaviours performed by calves while competing to reach the feeding table as calves competed for feed in the test. Data from analysed video recordings was transposed into an excel sheet for statistical analysis.

To account for the fact that recording was started before the test begun since the experimenters were still finishing with the set-up and had not left the barn, behavioural analysis of the video recording was timed and scheduled to start 180 seconds from the start of the video recording.

3.7. Statistical analysis

Statistical analyses were carried out using Minitab 18 software. Normal distribution was tested by the tolerance interval plots and conducting a 95% confidence interval and 95% of population interval. Breed was not uniformly distributed between groups, so it was not included in the statistical model. Therefore, only the variables that were assumed to be normally distributed were further analysed using Analysis of Variance (ANOVA) under the general linear model (GLMM) with a basic model consisting of two fixed factors: treatment, sex, and their interaction. Weight and age of the test calves were added to the basic model as covariates. The GLMM model was used to analyse the influence of the rearing systems (treatment factor with 2 levels: C and CCC), sex (factor with 2 levels: castrated male calves and heifer) and an interaction (factor with 2 levels: treatment and sex) of the calves as random variables considering the effect of their weights and age as covariates with all levels coded with values 0; 1. The fixed factors in the model were analysed in

response to behaviours recorded in the ethogram (Table 3) that were identified to be worth testing after exploring the data collected from all the observations and tests done with box plots.

To account for repeatability in data collected amongst calves, observations in grouped days were summed up to obtain a single result for each behaviour performed per calf. Analysed results are presented as means, standard error of the mean (SEM), F-values, and P-values with the level of significance set to $P < 0.05$. Individual play behaviours and focal scan behavioural observations were statistically analysed using the GLMM model in ANOVA. Social behavioural observations in this study were all analysed based on a descriptive statistical analysis tool. Breed as a factor was not analysed because it was too unevenly distributed between the two treatment groups to be included in the tests.

A T-test was done to identify the difference in the daily weight gain between the two treatments (CCC and C).

3.7.1. Focal scan observations

The focal scan observations entail instantaneous behaviours recorded as “I” in Table 3. These behaviours were recorded to be performed by an individual focal animal at a specific observation time for all 23 test calves.

On day 1, 2, and 3 (before mixing the calf groups), all data collected over a period of three days in a total time frame of 18hrs (6hrs/day) was summed for each behaviour to obtain one result for each calf. The resulting behavioural observation counts for each calf was analysed using the basic model described above.

On the 4th day which was the mixing day of the two calf groups, all observed behaviours explored for each behaviour on that day within the six-hour scheduled time were summed up to obtain one result for each calf. The single result obtained for each behaviour per calf was then analysed using the GLMM basic model.

On the last day of behavioural observation (5th day) which was the day after mixing, data collected over a period of 6 hours for all focal scan behaviours (Table 3) was as well summed up to obtain one result per behaviour for each calf. The results for each calf were analysed using the GLMM basic model.

For individual play behavioural analysis, the same protocol as described in behavioural observations for the five studied days was used to analyse all locomotor play behaviours (Table 3) observed during the five days in the study. Results for each behaviour per calf were then analysed as response variables in the described GLMM basic model.

3.7.2. Continuous observations

Continuous observations entailed all social behaviours recorded as “C” in Table 3. These behaviours were recorded continuously over a period of a 40-minute observation. The behaviours were recorded as being performed by an actor that initiated the behaviour and a receiver of the behaviour. If both animals initiated a behaviour, it was recorded as a mutual interaction between the animals for the behaviour observed. Given the fact that the social interaction observations were done on 23 calves that were observed continuously for five days without a comparative group of calves, a descriptive statistical analysis was done based on mean values and SEM obtained from observations done in three groups of days. The three groups of days consist of: the day before mixing (Day 1, 2 and 3), the mixing day (day 4) and the day after mixing (day 5).

Day before mixing

All social behavioural observations (Table 3) for the first three days of the study when calves were still in their treatment groups were combined into one result. Sum of each observed social behaviour (Table 3) based on the actor and receiver in each treatment group (C and CCC) and sex (castrated males and heifers) was recorded to obtain one result for better comparative and descriptive study analysis.

Mixing day

All data collected on day four (4th day) of social interaction for the day was summed up and a single value for each interaction was obtained based on actors and receivers in each treatment group and sex of calves interacting in each social behaviour observed and recorded.

Day after mixing

All data collected on the last behavioural observation day (5th day) was handled in such a way that, social interactions performed for each behaviour were summed up to obtain a total value of actors and receivers in each treatment group and sex of calves interacting in each social behaviour.

Percentage occurrence of the social behaviours observed to have the highest means based on the grouping within the three factors; treatment, sex and the interaction of actors and receivers within the treatments and sex was used to deduce results of the most performed social behaviours and by what treatment group and sex.

3.7.3. Runway test

Latency of test calf to re-unite with calf group was calculated as the time spent (<60s) from gate opening until calf were in close proximity (<1 m apart = zone 3) and relatively close proximity (<3 m apart = zone 2). Calves that were at a distant

proximity (>1 m apart = zone 1) including those that did not re-unite by <60 s, were considered not re-united.

Results on the latency of each calf to move from the starting line to the 5m and 2 m marks (Fig 6), mean duration and occurrence of calves in the three different zones, the number of vocalisations made by each test calf while in the start box and during the test, were all obtained from the behavioural analysis. The results were all analysed as responses with the described GLMM basic model consisting of the two fixed factors: treatment, sex, and their interaction. Movement patterns for the test calves during the test were also analysed together with the end zone based on their treatment group.

3.7.4. Avoidance distance test

ANOVA with the GLMM model of two fixed factors: Treatment (C; CCC), Sex (Heifer, castrated males) and their interaction were analysed in response to the distance moved by calves during the test.

3.7.5. Feed competition test

Behaviours recorded during the feed competition test were analysed using the GLMM model with the fixed factors consisting of Treatment (CCC and C), sex (castrated males and heifer) and an interaction of the two factors in response to data on eating behaviour at the feeding table by calves every after a 30 seconds time interval. The GLMM model was also used to analyse the described fixed factors in response to agonistic behaviours performed near the feeding area by calves while they competed to get feed. The ages and weights of the calves were included in the model as co-variates to determine their effects on the competitive behaviours performed during the test at the feeding area.

4. Study limitations

The calving rate at Lövsta farm and the distribution of calves between the two treatment groups resulted in an age difference of some weeks between the groups.

The age difference in calves between the two treatment groups in combination with weight gain or size of the calves could have influenced the competition effect and the agonistic behaviours performed during the Feed competition test.

Due to the difference in numbers of test calves between the two test groups that is C=13 and CCC= 10 both housed in a barn with space equally divided between the two groups. It resulted in a 1m² difference in the space allowance per calf between CCC and C treatment calf groups while housed in the barn in which the study was done as shown in figure 4. This could potentially be raised as a limiting factor in space allowance for play behaviour performed between the two groups. However, both groups received a space allowance per calf that surpasses the recommended 1.5 to 2m² space allowance per calf (Jordbruksverket, 2014). Therefore, space might not be considered as a limiting factor impacting on the social interaction at the initial stage since both groups performed play behaviours during the observations. However, one needs to be cautious, because we do not know the extent on how this affected their later behaviours.

While preparing to carry out the feed competition test, it was not possible to restrain the test calves in the facility given before putting the feed in the feeder. Therefore, the initial phase of test calves running to the feeder was not ensured.

5. Results

Analysed results are presented as means, standard error of the mean (SEM), F-values, and P-values with the level of significance set to $P < 0.05$.

All behavioural test results for individual play and focal scan behavioural observations were statistically analysed. Social behavioural observations in this study were all analysed based on a descriptive statistical analysis tool.

5.1.1. Focal scans

Before mixing the groups, the behaviours walking ($F=5.76$, $P=0.028$) and self-grooming ($F=8.55$, $P=0.009$) were performed more by the castrated male calves than the heifer calves. There was an effect of age ($F=4.92$, $P=0.041$) on the lying behaviour after mixing. However, no difference was found between treatments or sex in any of the observed behaviours on the mixing day and on the day after mixing the groups.

5.1.2. Individual play behaviours

Before mixing the groups, bucking ($F=5.91$, $P=0.026$) and jumping ($F=5.04$, $P=0.038$) behaviours were observed to be performed more by CCC calves than the C calves. There was an identified effect of age after mixing in butt fixture ($F=4.93$, $P=0.04$) and jumping ($F=6.60$, $P=0.02$) behaviours. However, there was no difference found between treatment and sex in any of the observed behaviours after mixing the groups.

5.1.3. Continuous observations

During the social observations, a descriptive statistical analysis was done which showed that there were generally more receivers during the observations than actors especially within the C group than in the CCC group (Figure 9). Higher social interaction on the mixing day were observed with more receivers in the C group than actors while in the CCC group, there were more actors than receivers which is a similar observation recorded the day after mixing but with a lower social interaction level (Figure 9).

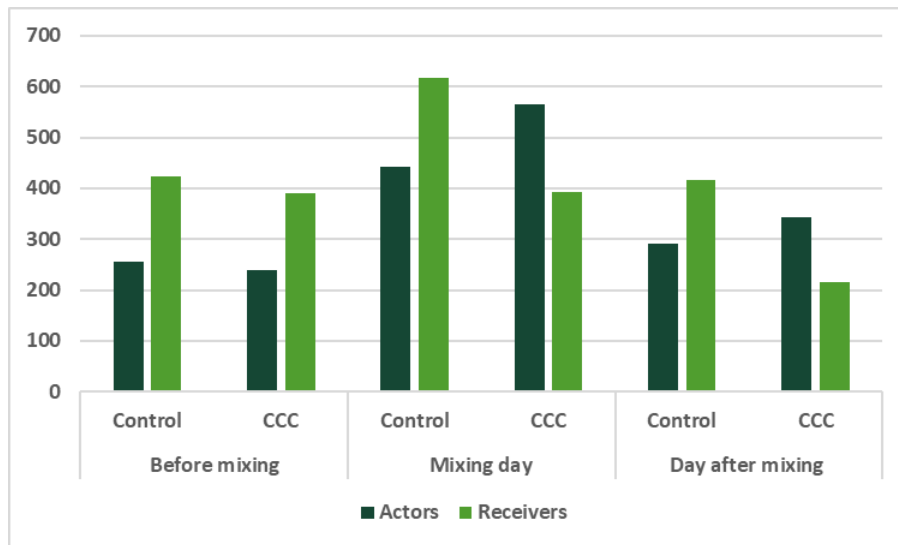


Figure 9. Sum of actors and receivers in all the observation days for social behaviours recorded in both the Control treatment and CCC treatment.

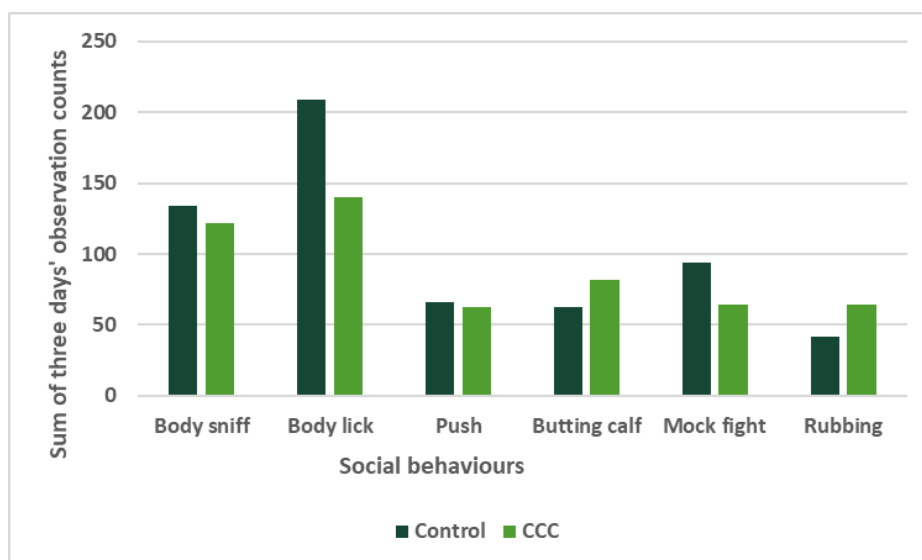


Figure 10. Sum of the first three days of social observations recorded to be performed by C and CCC calves before mixing the groups.

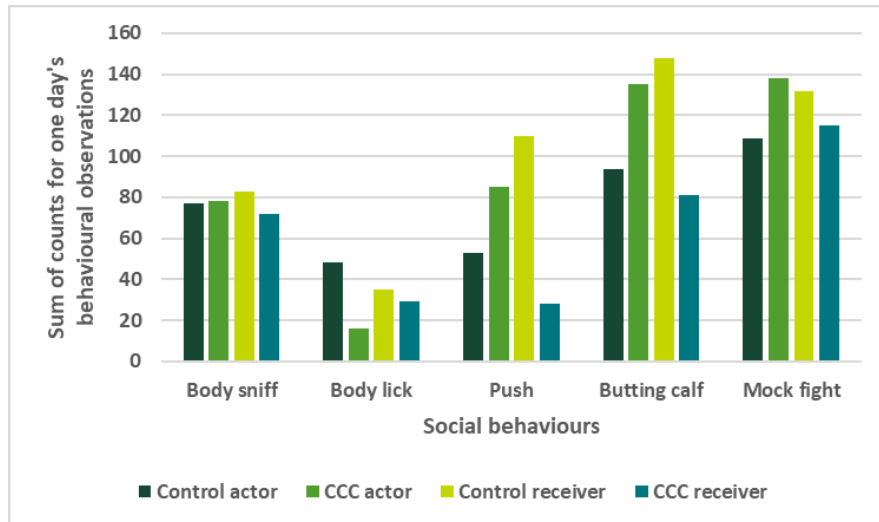


Figure 11. Sum per day of social behaviours recorded for actors and receivers in the newly formed mixed group containing both the Control treatment and CCC treatment calves on the mixing day.

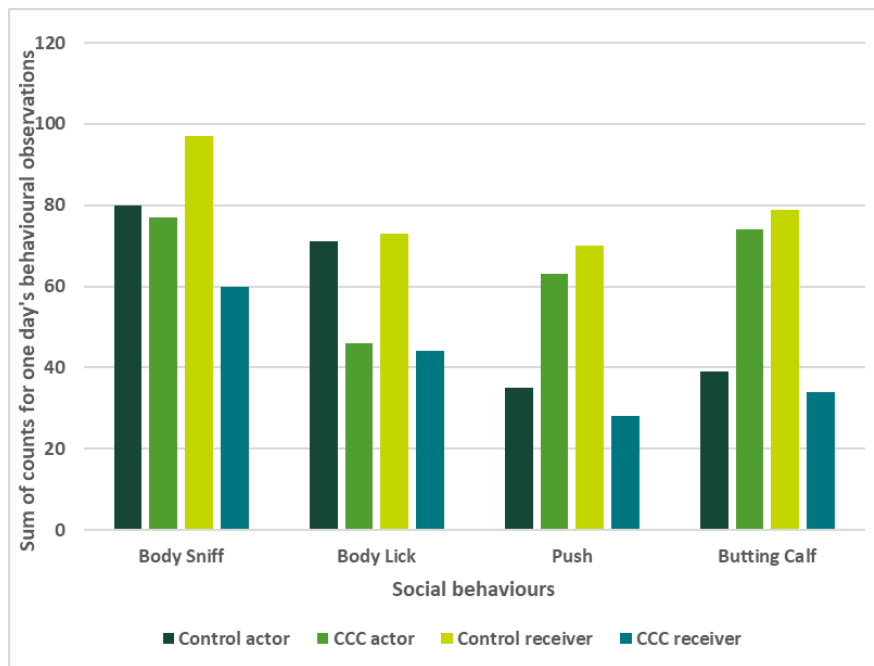


Figure 12. Sums for a day of social behaviours recorded for actors and receivers in both the Control treatment and CCC treatment while in the new mixed group on the day after mixing the groups.

5.2. Behavioural test

5.2.1. Avoidance distance test

There was a mean avoidance or flight distance of 0.9m for both CCC and C calves and with no statistical difference observed between the distance moved by the calves in the two test groups.

5.2.2. Runway test

There was an identified effect of age on latency from zone 1 to zone 2 ($F=7.77$, $P=0.016$) and the mean duration in zone 1 ($F=6.82$, $P=0.019$).

Five of the C calves and 7 of the CCC calves were observed to re-unite (i.e., calves in proximity; <1m apart and re-uniting in a latency <60s with group calves). Eight C calves and two CCC calves were never observed to re-unite (i.e. calves in proximity <1m apart or spent a latency beyond 60s to reach group calves and those that never crossed *) as shown in Table 4.

The latency to re-unite was highest in the CCC calves and lowest in the C calves as shown in Table 4.

Table 4. Latencies of each test calf to re-unite with group and their end zones during the runway test.

Treatment	Mean Lat.Zn1-Zn2	Mean Lat.Zn1-Zn3
C calves	25.5	34.9
CCC calves	33.1	22.8

*Calves that did not cross the 5m and 2m mark hence did not reach the next zones.

†Missing data for the calf due to battery shut down and human error.

Heifers had a higher latency from zone 1 to zone 3 ($F=5.57$, $P=0.031$) with a high interaction in the latency from zone 1 to zone 3 between treatment and sex ($F=5.71$, $P=0.029$). The interaction involved; castrated males having a slightly higher latency from Zone 1 to zone 3 (Mean=35.5) from the heifers (Mean=12.6) in the CCC treatment while heifers had a higher latency from zone 1 to zone 3 (Mean=54) than the castrated males (Mean=12.67) in C group.

Movement patterns in the runway test

Control group

Of the 13 C calves, four calves (2 castrated males and 2 heifers) remained in Zone1 the entire test time without crossing to another zone. The remaining nine calves crossed from Zone1 to Zone2 and 3 (Fig 6) with different movement patterns.

Zone1 had the highest mean duration (126 seconds) followed by Zone3 (19 seconds) and lastly Zone2 (12 seconds)

Nine calves (4 castrated males and 5 heifers) had zone1 as their end zone, only one heifer calf ended in Zone2 and three calves (2 castrated males and 1 heifer) had Zone3 as their end zone by the end of the test.

CCC treatment group

One calf was not recorded, therefore, of the nine recorded CCC test calves, all the nine calves crossed from Zone 1 to Zone 2 and Zone 3 within the runway test.

Zone 3 had the highest average mean duration (68 seconds) followed by Zone 1 (51 seconds) and lastly zone 2 (24 seconds).

Three calves (2 castrated males and 1 heifer) had Zone 1 as their end zone, three calves (2 castrated males and 1 heifer) had Zone 2 as their end zone and three calves (all heifers) had zone 3 as their end zone at the end of the test.

5.2.3. Feed competition test

There was no significant difference in the eating behaviour between CCC and C calves on the feeding table during the test.

Chin pressing ($F=7.40$, $P=0.022$) behaviour was predominantly performed by CCC calves around the feeder while competing for feed. Furthermore, a high interaction was observed in rubbing ($F=7.24$, $P=0.023$) and chin pressing ($F=5.91$, $P=0.035$) behaviours between treatment and sex. The interaction involved castrated males performing more rubbing behaviour (Mean=2.00, SEM=1.0) than heifers (mean=0.455, SEM=0.21) in the CCC treatment calves while heifers performed more rubbing behaviour (Mean=0.33) than castrated males (Mean=0) in the C group calves.

Chin pressing behaviour was predominantly performed by CCC calves (mean=2.13; SEM= 0.58) compared to C calves (mean=0.50; SEM=0.38, $F=17.21$, $P=0.002$). There was a significant interaction between treatment and sex ($F = 5.51$, $P = 0.039$) around the feeder with CCC castrated male calves (mean = 2.20, SEM = 0.92) interacting more than the heifers and C group calves while competing for feed.

During the test, 11 out of the 13 C calves, competed and appeared next to the feeder. Seven of the eleven C calves appeared more consistently at the feeder throughout the experiment. For the CCC calves, nine out of the 10 CCC calves, competed for feed throughout the entire test around the feeder. Out of the nine CCC calves, eight of them constantly competed and appeared more consistently around the feeder throughout the experiment.

6. Discussion

6.1. Affiliative and agonistic behaviours observed

With the identified difference in age, weight gain and stocking rate between the two test groups, results obtained from statistically analysed behavioural observations showed no effect of weight on the behaviours given that CCC calves gained more weight than C calves. However, age could have influenced some locomotor and individual play behaviours in the groups and during the runway sociality test with the CCC calves having been slightly older than the C calves.

The individual play behaviours and descriptively analysed social behaviours show potentially more social sniffing and licking (affiliative) behaviours in C calves but lower agonistic play behaviours. CCC calves performed more agonistic play behaviours and potentially higher social behavioural interaction mainly involving pushing, butting, mocking, and rubbing. Results could be related with earlier studies that show that more affiliative behaviours or non-agonistic and fewer agonistic behaviour encounters are associated with calves previously kept in individual crates, pens or hutches compared to calves reared with social contact or in groups (Veissier et al., 1994). From the observed results on the mixing day and the day after mixing the groups which resulted into a single combined new group; the social motivation to initiate social interaction was observed to be higher in CCC calves than in C calves with the interaction involving both familiar and unfamiliar mates. In this study, CCC calves showed the ability to socially interact with both familiar and unfamiliar mates unlike in other research (Duve and Jensen, 2011) that showed that calves form stronger social bonds to peers that they have had full social contact with either from birth or from three weeks of age as seen in C group calves. The results showed that CCC calves potentially have higher dominance as actors and might have more developed social behaviours which could enable them to behave better in a new group than C calves that showed more submissive behaviours as receivers in most social interactions.

6.1.1. Locomotive and individual play behaviours

Having statistically analysed all individual play behaviours done through the focal scan observations, walking and self-grooming behaviours were both identified to be highly performed by heifers than castrated male calves. Self-grooming is considered a non-agonistic behaviour while walking is a state and posture that may be motivated by movement of mates or due to a factor of age (Fraser and Broom., 1997). Further research could be done on the relationship between activity in movements or state posture with age as the development of hormones and maturity of calves for example bull calves, could contribute to their active movement in search of a female partner or mate.

CCC calves performed more bucking and jumping play behaviours than the C calves. Agonistic behaviours like bucking and mock fighting are termed as play behaviours in calves. These behaviours are expected to be exhibited by calves socially grouped earlier in life and animals that have been raised with space and social contact (Jensen et al., 1999). The space and social contact increasingly stimulate play and social behaviours (Jensen et al., 1999) similarly observed in CCC calves. This could indicate that the early social life experience of the CCC calves stimulated the development of individual and social play behaviours plus social interaction traits that they depicted with both familiar and unfamiliar mates when grouped.

6.1.2. Social interactions

All social behavioural interactions were descriptively analysed in this study.

On the mixing day when a new group was formed, higher sums of social interactions were recorded than before mixing the groups in which case, C calves were mainly receivers than actors while CCC calves mainly initiated behavioural interactions as actors in the social interactions than receivers. The trend observed on the day after mixing also involved the CCC calves being the main actors in most behaviours while the C calves were the main receivers in all behaviours. The results show that CCC calves that had body and social contact at an early stage in life could be more explorative and might have more developed social behaviours like locomotor play and play fighting which are reported to usually begin at two weeks of age (Waldau, 2017). This could have influenced the social motivation of CCC calves to interact by initiating social interactions within the new group even in the presence of unfamiliar mates. C calves however mainly initiated social affiliative behaviours and their interactions mainly involved them being the main receivers than actors. The results potentially show how CCC calves set up their hierarchy or dominance in a group by easily socializing and blending into a new group with unfamiliar mates. The results are similar to a study done by Broom & Leaver (1978) which showed that group reared calves were observed to initiate more contact and interactions and to be more dominant over individually raised calves when they are

put together in one pen. However, the C calves might be highly submissive mates when grouped with unfamiliar mates as shown in the social interactions results were, they were mainly more of receivers than initiators of behaviours especially when they interacted in the new group. Results tally with a study done by Broom & Leaver (1978) that individually housed calves seemed to be highly incompetent in social encounters. This could potentially mean that early social environment, social contact, and interactions could greatly contribute to the development of social behaviours and retaining of the sociability traits in dairy calves.

Before mixing

Body sniffing and licking, pushing, butting calf and mock fighting were the behaviours performed predominantly in the social interaction between the calves while still housed in their treatment groups. C calves were observed to socially body sniffed and licked more than the CCC calves which behaviours are described as affiliative social interactions that reinforce a social bond within a group. Body sniffing and licking are considered as synchronized affiliative behaviours that happen with a lick following a sniff common in cattle as gregarious animals (Šárová et al, 2007). These affiliative behaviours could have manifested with their mates either naturally to mutually maintain hygiene/groom each other or in a natural bid to socially integrate (Jensen et al., 1999). Duve and Jensen (2012)'s report states that, individual housing not only hinders the calves in performing certain aspects of their social behaviour repertoire but also limits the time spent on social behaviours that can be performed through the bars. This could mean that the C calves' early experience and restriction in individual pens in the first eight weeks could have affected the locomotor, exploratory and social behaviours of the calves in several ways like hindering or reducing the development of some social behaviours. Results obtained relate to Jensen et al., (1999) and Waldau, (2017) reports that an early social experience through visual, auditory, and tactile contact but not body contact reinforces the calves' social bond through the affiliative behaviours performed. When the C and CCC calves were mixed to form a new group, there was a descriptively observed preference of C calves for familiar mates over unfamiliar ones. From the descriptive analysis, C calves performed more pushing and mock fighting while in their treatment groups than CCC calves, and those behaviours have been suggested to be friendly agonistic behaviours, which strengthen the social cohesion in cattle herds (Reinhardt et al., 1986). They may also be regarded as interactive play behaviours between calves (Reinhardt et al., 1978) hence showing that earlier grouping of individually housed calves with their mates could influence the learning of some social skills.

CCC calves performed more butting and rubbing behaviour than the C calves. This could mean that CCC calves initiated agonistic play and interactive behaviours more easily in a social environment. CCC calves depicted a desire for

companionship with neighbour attachment through rubbing. Butting is considered an aggressive behaviour that increases with age and may be involved in the development of a social hierarchy among calves (Reinhardt et al., 1978). CCC calves could be identified to show superiority or potentially be higher hierarchy achievers which could be a good characteristic for grouping of dairy calves in their later life and when socially challenged in a new group. This could be because, mothered calves have been reported to be more active and to dominate the ones separated from their mothers (Schleyer and Karminsky, 1997; Veissier and Le Neindre, 1989).

Mixing day

The CCC calves were predominantly the main actors and initiators of all the predominantly performed social behaviours (body sniff, mock fighting, butting calf and pushing) while the C calves were the main receivers. This might show that individually housed calves might tend to obtain a lower rank in the hierarchy after grouping compared with calves with previous social experiences (Veissier et al., 1994). Pushing, butting and mock fighting are identified as features of social reactivity and variants of play in young animals with developed social interactions due to the early life social experience and contact (Reinhardt and Reinhardt, 1982). This could mean that CCC calves that predominantly performed these agonistic play behaviours might have a higher rank and a more developed social interactive ability, motivation, and sociability traits from their early life experience which potentially benefited their later life.

Heifers were predominantly the main actors in all social behavioural interactions stated above with fellow heifers being the main receivers in all the interactions.

Day after mixing

On the day after mixing, body sniff, body licking, butting calf and pushing in that order were the most performed social interaction between the two treatment groups of calves. Social body sniff and lick were mainly initiated and performed by C calves and similarly, all the social interactions were mainly received by the C calves on that day during the study. Initially before mixing the groups, C calves mainly performed the sniffing and licking behaviour more than the other social behaviours. The results obtained on the day after mixing could therefore be a reinforcement of what Fraser and Broom (1997) stated that social factors might have a role in the development of behaviours based on the fact that once an animal in a group learnt any behaviour, others in the group are likely to learn to do the behaviour too. Hence, the C calves learning, maintaining, and predominantly performing the synchronized social sniffing and licking as earlier performed before mixing could have been well developed behaviours in their group which they mainly reciprocated with their mates.

Since the CCC calves initiated more social behaviours after mixing than the C calves, one could say that they might be more sociable and more prone to socialise and initiate an interaction even with new mates in a group. Butting and pushing are identified as agonistic social play behaviours in young animals. As seen in my descriptive study results, these behaviours were performed more by CCC calves than C calves. Interaction was not only with their fellow CCC mates, but also with the C calves that were the most prominent receivers of interactions in the observations. This could prove the potential positive influence of early learning, development, and motivation of behaviours in CCC calves attained in their tender age while growing with their dams, other cows, and peers.

Social interactions within the sex of the calves were high with inter-sexual social interaction between the heifers and castrated male calves being the highest followed by the heifer-to-heifer social interactions. In all highly performed social behaviours during the interactions, heifers were identified to be the main actors performing the behaviours on fellow heifers. The observation can be supported by a study that was done on young dairy heifers by Broom and Leaver (1978), which showed that associations were more likely to happen among heifers reared together as calves than others reared differently and of different age groups.

6.2. Socialization

6.2.1. Social motivation and sociability

The CCC calves expressed a high social motivation to re-unite with their groupmates after separation shown as shorter latencies to approach the group. CCC calves' short latencies between zone 1 and zone 3 shows their sociability traits unlike the C group that had high latencies with eight of the C calves failing to re-unite including four calves that never left the starting area (Zone 1 which was close to the gate). The consistency depicted by CCC calves crossing to different zones as observed in the movement patterns, and the high number of CCC calves that ended in Zone 3 relative to the performance of C calves shows a potential higher motivation in CCC calves to re-unite with their group than the C calves. The test results of a higher social motivation in the CCC calves to re-unite with the group mates exhibited by the lower latency to reach the group and longer durations spent near the group; potentially show the extent to which CCC calves need social companionship and how much they want to be close to their group mates than C calves as in a paper by Keeling and Gonyou (2001).

Using a descriptive presentation of the results obtained in the study carried out, all CCC calves vocalised at least once while in isolation and during the test and the highest vocalizations were recorded from the CCC treatment group. On the other hand, for the C calves, 5 of the calves neither vocalized while in isolation in the

start box nor during the test. Vocalizations from CCC calves was higher and more frequent than for the C calves during the test arena while both in isolation in the start box and during the test. Fraser and Broom (1997), states that vocalization is a crucial communication behaviour or language for animals, and it can be a sign of alarm for distress. Therefore, these vocalizations could most likely be an indicator of separation distress, analogous to the vocal responses of the calves after separation from the group (Flower and Weary, 2001). These results could suggest that calves reared in social contact with their dams, other older cows and peers form stronger social bonds and develop sociability traits (need of companionship) hence the high vocalization when separated from their peers or group.

6.2.2. Sociality

The aim of this test was to test for the calves' docility in the presence of human handlers through the flight distance moved by calves in the two treatment groups to assess the calf's fear towards humans or willingness to interact and get close to the handler. Docility has a lot to do with sociality of an animal in the presence of humans and it is the primary and essential trait for domestication (Kretchmer and Fox, 1975).

Results obtained, showed that there was no difference in the distances moved between the C and CCC calves with an average flight distance of 0.9m moved for both treatment groups from the handler. This indicates that the animals in both groups are docile and can easily interact, socialize, and be well handled by humans. As stated by Hale, (1961), a short flight distance to humans and the ability not to be easily disturbed by humans or sudden changes in environment are favourable behavioural characteristics that favour domestication of a species. However, it should also be noted that results could have been affected by the fact that all calves were subjected to a daily interaction with a human handler to avoid aggressive behaviours while being raised so that they are habituated to human handling.

6.2.3. Social skills and sociability traits.

Agonistic social interactions are those related to conflict and competition, including threat or aggression ("C" behaviours described in Table 3), avoidance and submissive behaviours (de Oliveira et al., 2020; Boulssou et al., 2001; Mills et al., 2010). All social interactions observed were statistically analysed.

Competition for feed at the feeder was not clearly identified because calves were not locked up before the start of the test and during the placement of feed. Therefore, calves already positioned at the feeding table had the opportunity to feed before the test begun and hindered the initial competitive behaviours of calves moving towards the feeding table. However, Syme (1974) points out that competition for food item is often resolved by the faster mover acquiring it.

Therefore, the calves identified to be at the feeding table can most likely be equipped with a fast acting and moving ability to the feeding table rather than fighting to compete for feed.

Mounting, chin press and rubbing behaviours were predominantly observed to be performed by the CCC calves around the feeder towards other calves as they competed for feed. This could mean that social contact can influence the competitiveness of the dairy calves. Mounting, Chin press and rubbing behaviours could on one hand be identified as social agonistic behaviours but also on the other hand as threat behaviours as described in Fraser and Broom (1997). High agonistic behaviours performed by CCC calves in the test therefore affirm the high rank and dominance of the CCC calves and their ability to exert threat behaviours in a challenged environment to acquire feed. This could imply that CCC calves can learn the signals, behave better, and explore an environment faster than C calves in a group.

However, the number of feeding places, the size of the feeding area, the feed barrier design and the amount and type of food influences the motivation to feed and the level of competition (Metz, 1983). Therefore, the feed competition might have greatly been affected or influenced by the presence of feed on only one feeding area that was best known and commonly used by the C calves. A neutral and new feeding area could have been created to purely test the calves based on neutral standardized factors that are not common to both treatments in the novel environment to enhance competition.

Olofsson, (1999) wrote that reducing the number of available feeding places increases the frequency of displacements at the feeding station and the width of the feeding place will also influence the level of competition (Zeeb et al., 1988). Therefore, the size and the feed placed in the feeding area during the test could have been more than required in relation to the number of test calves which in turn could be a reason that hindered proper competition and behavioural motivation in the test.

7. Conclusion

This pilot study suggests that CCC calves might potentially perform more play and agonistic behaviours, socialize, and interact more than C calves. CCC calves might additionally express higher sociability traits and normal calf behaviours than C calves especially when socially challenged in a new group. CCC calves might have a higher ability to learn and develop social, explorative, and natural behaviours. The study could in part potentially confirm that early socialization of a calf with dams and peers during the first months of life could contribute to the development of sociality traits in dairy calves. It further might contribute to the later social behaviours of the calves when separated and re-grouped with unfamiliar mates.

7.1. Recommendations

A broader and wider multicentric study over a longer period involving different breeds needs to be undertaken. This is to evaluate the potential effects of early socialization to genetically inherited behaviours from different breeds on the social behaviours in the CCC calves in their first months of life and on their later lives.

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Appendix 1

Experimental Ids of Calves								
No.	Key	Tag Id	Colour	Colour initial	Type	Breed	Sex	Treatment
1	A	2266	Orange + Blue	OB	Brown	Srb	Heifer	Control
2	B	2267	Green	G	Holstein	Sh	Heifer	Control
3	C	8211	Red	R	Brown	Srb	Bull calf	Control
4	D	8207	Cream + Orange	CO	Brown	Srb	Bull calf	Control
5	E	2269	Double Cream	CC	Brown	Srb	Heifer	Control
6	F	2272	Pink	P	Holstein	Sh	Heifer	Control
7	G	2264	Blue + Blue + Pink	BBG	Brown	Srb	Heifer	Control
8	H	8208	Yellow	Y	Brown	Srb	Bull calf	Control
9	I	8205	Blue + Green	BG	Holstein	Sh	Bull calf	Control
10	J	2265	White	W	Holstein	Sh	Heifer	Control
11	K	2275	Orange	O	Holstein	Srb	Heifer	Control
12	L	8206	Blue	B	Holstein	Sh	Bull calf	Control
13	M	8214	Blue + Yellow	BY	Brown	Srb	Bull calf	Control
14	N	2258	Pink + Orange	PO	Brown	Srb	Heifer	CCC
15	O	2259	Red + Yellow	RY	Brown	Srb	Heifer	CCC
16	P	8201	Double Orange	OO	Holstein	Sh	Bull calf	CCC
17	Q	8198	Double Blue	BB	Holstein	Sh	Bull calf	CCC
18	R	8204	Yellow + Green	YG	Holstein	Sh	Bull calf	CCC
19	S	8199	Pink + Green	PG	Holstein	Sh	Bull calf	CCC
20	T	2262	-	-	Brown	Srb	Heifer	CCC
21	U	2261	Green + Orange	GO	Brown	Srb	Heifer	CCC
22	V	2268	Cream + Pink	CP	Brown	Srb	Heifer	CCC
23	W	2263	Double Green	GG	Brown	Srb	Heifer	CCC

observer 1				observer 2			
start time	end time	duty	group	start time	end time	duty	group
09:00	09:10	preparing material/cameras	1	09:00	09:10	preparing material/cameras	2
09:10	09:15	focal scan	1	09:10	09:15	focal scan	2
09:15	09:55	social interactions	1	09:15	09:55	social interactions	2
09:55	10:00	focal scan	1	09:55	10:00	focal scan	2
break (20')				break (20')			
10:20	10:25	focal scan	1	10:20	10:25	focal scan	2
10:30	11:10	social interactions	1	10:30	11:10	social interactions	2
11:10	11:15	focal scan	1	11:10	11:15	focal scan	2
11:15	11:55	social interactions	1	11:15	11:55	social interactions	2
lunch (60')				lunch (60')			
13:00	13:05	focal scan	2	13:00	13:05	focal scan	1
13:05	13:45	social interactions	2	13:05	13:45	social interactions	1
13:45	13:50	focal scan	2	13:45	13:50	focal scan	1
13:50	14:30	social interactions	2	13:50	14:30	social interactions	1
14:35	14:40	focal scan	2	14:35	14:40	focal scan	1
break (20')				break (20')			
15:00	15:05	focal scan	2	15:00	15:05	focal scan	1
15:05	15:45	social interactions	2	15:05	15:45	social interactions	1
15:45	15:50	focal scan	2	15:45	15:50	focal scan	1

observer 1				observer 2			
start time	end time	duty	group	start time	end time	duty	group
09:00	09:15	fix cameras/material	1	09:00	09:15	fix cameras/material	2
09:20	09:40	remove wall	1	09:20	09:40	remove wall	2
09:40	10:40	social interactions	1	09:40	10:40	social interactions	2
10:40	10:45	focal scan	1	10:40	10:50	focal scan	2
break (20')				break (20')			
11:05	11:10	focal scan	1	11:05	11:10	focal scan	2
11:10	11:50	social interactions	1	11:10	11:50	social interactions	2
11:50	11:55	focal scan	1	11:50	11:55	focal scan	2
lunch (60')				lunch (60')			
13:00	13:05	focal scan	2	13:00	13:05	focal scan	1
13:05	13:45	social interactions	2	13:05	13:45	social interactions	1
13:45	13:50	focal scan	2	13:45	13:50	focal scan	1
13:50	14:30	social interactions	2	13:50	14:30	social interactions	1
14:35	14:40	focal scan	2	14:35	14:40	focal scan	1
break (20')				break (20')			
15:00	15:05	focal scan	2	15:00	15:05	focal scan	1
15:05	15:45	social interactions	2	15:05	15:45	social interactions	1
15:45	15:50	focal scan	2	15:45	15:50	focal scan	1
Day 2 follow the schedule of social interactions							